[3]*Ghent University Hospital, Radiation Oncology and Experimental Cancer Research, Ghent, Belgium*
[4]*University Hospital Leuven, Abdominal Surgery, Leuven, Belgium*
[5]*University Hospital Leuven, Digestive Oncology, Leuven, Belgium*
[6]*University Hospital Leuven, Radiology, Leuven, Belgium*
[7]*University Hospital Leuven, Pathology, Leuven, Belgium*
[8]*University Hospital Leuven, Nuclear Medicine, Leuven, Belgium*
[9]*Stanford University, Medicine- Division of Biomedical Informatics, Stanford, USA*

Purpose or Objective: Selecting good responders after chemoradiation (CRT) for locally advanced rectal cancer (LARC) could lead to the omission of total mesorectal excision (TME) in patients with pathologic complete response (pCR). In the current study, we assessed the value of several blood biomarkers associated with the fibrotic response to CRT (IGF-1, IGFBP-2, HGF & GDF-15) as markers for general fibro-inflammatory response and as tumor response predictors in a group of 80 patients.

Material and Methods: ELISA analysis of IGF-1, IGFBP-2, HGF and GDF-15 was conducted on prospectively collected serum samples of 80 LARC patients on 3 time points (before, during, after CRT). The fibro-inflammatory response was scored on H&E sections of the resection specimen. Changes in concentration were analysed using a Kruskal-Wallis test. Correlation of concentration at each time point and the difference between these time points (Δ) with fibro-inflammatory response and tumor response (pCR and ypT0-1) were assessed using a Mann-Whitney-U test.

Results: Higher Growth Differentiation Factor 15 (GDF-15) concentration before CRT correlated with the presence of a fibro-inflammatory response ($p = 0.04$), but was not observed for the other proteins nor for GDF-15 at other time points.
General increase in GDF-15 concentration during treatment (median 0.81 ng/ml before, 2.16 ng/ml during, 2.37 ng/ml after CRT; $p < 0.0001$) was measured (Figure 1). Although no significant general concentration changes occurred for IGF-1, IGFBP-2 or HGF, we did find a correlation between the variation in expression of IGFBP-2 during treatment (ΔIGFBP-2 TP3-TP2) with tumor response (pCR $p = 0.02$; ypT0-1 $p = 0.02$). Other proteins did not correlate with tumor response.

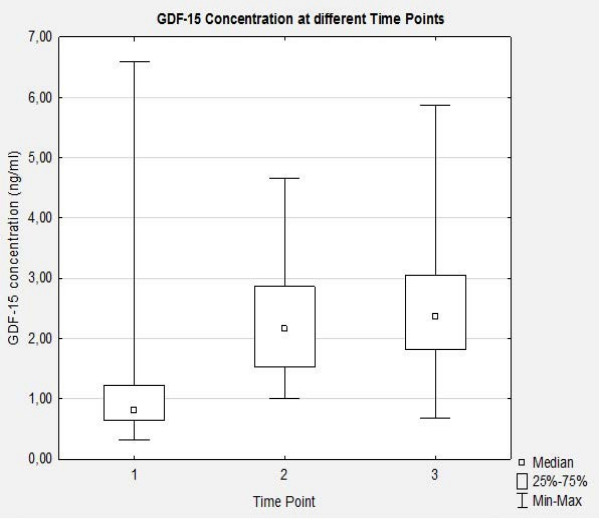

Figure 1. GDF-15 Concentration at different time points (TP) during treatment. (TP1: before CRT; TP2: during CRT; TP3: after CRT)

Conclusion: GDF-15 serum concentration increases during CRT for LARC and a higher concentration measured before start of treatment is correlated with the presence of a fibro-inflammatory response. These results suggest that GDF-15 could be used as an early predictor of fibro-inflammatory response and thereby indirectly as predictor for disease-free survival. This will be evaluated when follow-up data are available for this patient cohort.
The correlation of variation in expression of IGFBP-2 with tumor response (pCR and ypT0-1) opens a novel possibility for selecting good responders to CRT. We aim to combine these findings with imaging analyses (DW-MRI, PET) at different time points during treatment to develop a predictive model for selecting LARC patients in whom surgery could be omitted.

EP-2056
Preclinical investigation of hypoxia induced genes in different prostate cancer cell lines.
T. Wittenborn[1], S. Nielsen[1], M. Busk[1], M.R. Horsman[1], J. Overgaard[1], J. Alsner[1], B.S. Sørensen[1]
[1]*Aarhus University Hospital, Department of Experimental Clinical Oncology, Aarhus, Denmark*

Purpose or Objective: Hypoxia is a common feature in prostate cancer and is known to reduce the response to radiotherapy. Hypoxic modifiers can to a large extent overcome these obstacles, and a proper classification of tumors into hypoxic and non-hypoxic fractions is necessary. Previously our department has developed a gene profile consisting of 15 genes, which demonstrated prognostic and predictive impact for hypoxic modification in head and neck squamous cell carcinomas (HNSCC). In the current study we investigated the 15 gene profile in different prostate cancer cell lines.

Material and Methods: For the in vitro experiments the prostate cancer cell lines investigated were PC-3, DU-145, and LNCaP. Cell lines were cultured under normoxic (21% O2) or hypoxic conditions (0% O2) for 24 hours, totRNA was extracted and gene expression levels measured by qPCR. Individual reference genes were selected (PSMC4, TBP, NDFIP1) and applied in the normalization of the relative expression levels, together with the reference genes previously used in the HNSCC study. For in vivo experiments, the PC3 cell line was inoculated on the flank of female NMRI nu/nu mice, whereas the LNCaP and DU-145 cell lines were inoculated on the flank of severely immunocompromised CIEA/NOG mice. Two hypoxia-sensitive tracers (18F-FAZA and Pimonidazole) were administered in order to determine hypoxic and non-hypoxic regions on excised tumor sections. These regions were isolated by laser-assisted microdissection, after which totRNA was extracted and gene expression levels measured by qPCR.

Results: In the in vitro experiments, all prostate cancer cell lines had 14 of the 15 genes induced by hypoxia. The only discrepancy was ALDOA, which was not upregulated in the hypoxic cells. In vivo experiments are still ongoing but preliminary results from PC3 xenografts have been produced. These show a hypoxia induced upregulation in 10 out of the 15 genes, of which 4 were significantly upregulated (ADM, ANKRD37, FAM162A, and LOX).

Conclusion: In this study we investigated the 15 gene hypoxic profile in three different prostate cancer cell lines. A hypoxia dependent induction of genes was observed in both in vitro and in vivo experiments. From the performed experiments, and looking only at oxygen dependency, it appears that the gene profile could be suitable for prostate cancers as well as HNSCC.

EP-2057
Radiotoxicity prediction by gene expression profiling when simulating therapy in matched fibroblasts
M.A. Schirmer[1], C.P.N. Mergler[1], L.H. Droege[1], M. Guhlich[1], J. Gaedcke[2], M. Ghadimi[2], M. Rave-Fränk[1]
[1]*Klinikum der Universität Göttingen, Radiotherapy and Radiation Oncology, Göttingen, Germany*
[2]*Klinikum der Universität Göttingen, General and Visceral Surgery, Göttingen, Germany*

Purpose or Objective: Acute radiotoxicity might put a vital threat to the patient and may require interruption or